A request for a one-month extension of time to respond is included herewith along with the required fee. This one-month extension will bring the due date to November 7, 1998, which is within the six-month statutory period. Should such request or fee be deficient or absent, consider this paragraph such a request and authorization to withdraw the appropriate fee under 37 C.F.R. §§ 1.16 to 1.21 from Arnold, White & Durkee Deposit Account No. 01-2508/UTXC:504/STA.

Reconsideration of the application is respectfully requested.

I. AMENDMENT

Please make the following amendments:

In the Claims:

Please amend the claims as follows:

1. (Thrice Amended) A composition comprising a first polynucleotide that hybridizes to a second, Bcl-2-encoding polynucleotide under intracellular conditions and a neutral lipid associated with said first polynucleotide to form a Bcl-2 polynucleotide/neutral lipid association.

21. (Amended) A method of inhibiting proliferation of a Bcl-2-associated disease cell having a t(14;18) translocation comprising:

(a) obtaining an oligonucleotide [nucleotide] of from about 8 to about 50 bases and complementary to at least 8 consecutive bases of the translation initiation site of Bcl-2 mRNA;

(b) mixing the oligonucleotide with a neutral lipid to form a neutral oligonucleotide/lipid association; and

(c) administering said association to said Bcl-2-associated disease cell to inhibit the proliferation of said disease cell.

Please add the following claims:

--38. The composition of claim 1, wherein said first polynucleotide is a P-ethoxy oligonucleotide.

39. The composition of claim 5, wherein said liposome consists essentially of neutral lipids.

40. The composition of claim 9, comprising a liposome formed from said neutral lipid.

41. The composition association of claim 40, wherein said liposome consists essentially of neutral lipids.

42. The composition of claim 9, wherein said first polynucleotide is a P-ethoxy oligonucleotide.

43. The method of claim 10, wherein said first polynucleotide is a P-ethoxy oligonucleotide.

44. The method of claim 14, wherein said liposome consists essentially of neutral lipids.

45. The method of claim 21, wherein said first oligonucleotide is a P-ethoxy oligonucleotide.

46. The method of claim 24, wherein said liposome consists essentially of neutral lipids.

47. The neutral lipid oligonucleotide association of claim 31, wherein said first oligonucleotide is a P-ethoxy oligonucleotide.

48. The neutral lipid oligonucleotide association of claim 33, wherein said liposome consists essentially of neutral lipids.

49. The composition of claim 37, comprising a liposome formed from the lipid.

50. The composition of claim 49, wherein said liposome consists essentially of neutral lipids.

51. The composition of claim 37, wherein said oligonucleotide is a P-ethoxy oligonucleotide.

52. A composition comprising a first polynucleotide that hybridizes to a second, Bcl-2-encoding polynucleotide under intracellular conditions and a primary phosphatide associated with said first polynucleotide, wherein said primary phosphatide is a neutral lipid.

53. The composition of claim 52, comprising a liposome formed from the primary phosphatide.

54. The composition of claim 53, wherein said liposome consists essentially of neutral lipids.

55. The composition association of claim 52, wherein said first polynucleotide is a P-ethoxy oligonucleotide.--

II. RESPONSE TO OFFICE ACTION

A. Status of the Claims

Claims 1 and 21 have been amended to improve their clarity. No claims have been canceled and claims 38-55 have been added. Claims 1-55 are therefore pending.

For the convenience of the Examiner, a copy of the pending claims is attached hereto as **Exhibit A**.

B. Support for the Revision

Claim 1 has been amended to clarify that the Bcl-2 polynucleotide and neutral lipid "form a Bcl-2 polynucleotide/neutral lipid association." Support for this amendment can be found in the specification and claims as filed, at least at page 6, lines 16-19; page 23, lines 4-5; and originally filed claim 10.

Claims 21 has been clarified to replace the term "oligonucleotide nucleotide" with the term "oligonucleotide", which is supported by the specification and claims as filed, at least at page 13, lines 10-12:

> As stated above, although the antisense sequences may be full length genomic or cDNA copies, or large fragments thereof, they also may be shorter fragments, or 'oligonucleotides,' defined herein as polynucleotides of 50 or less bases.

New claims 38-55 have been added for the Examiner's review. Any additional fees necessitated by the presently added claims should be deducted from Applicants' representative's Deposit Account No. 01-2508/UTXC:504/STA.

Claims 38, 42, 43, 45, 47, 51 and 55 have been added to describe embodiments of the invention wherein the polynucleotide or oligonucleotide is a P-ethoxy oligonucleotide. Support for these claims may be found throughout the specification and claims as filed. Particular written support is found at least at page 25, lines 13-15:

> P-ethoxy oligonucleotides, nucleases resistant analogues of phosphodiesters, are preferred because they are stable in serum and effectively transported into the cellular cytoplasm.

Additional written support for "P-ethoxy oligonucleotides" can be found at page 33, lines 5-6, and page 33, lines 13-14.

Claims 39, 41, 44, 46, 48, 50 and 54 have been added to describe embodiments of the invention wherein the liposome "consists essentially of neutral lipids." Support for these claims may be found throughout the specification and claims as filed. Particular written support includes the teaching of neutral lipids used in the preparation of liposomes, at page 23, lines 4-5:

> Phospholipids are used for preparing the liposomes according to the present invention and can carry a net positive charge, a net negative charge or are neutral.

The specification teaches that other lipids, including charged lipids, may comprise a liposome, as described at page 23, lines 4-20. The specification also teaches that liposomes may have other lipids by its definition of the "primary phosphatide" constituting 50% or more of a liposomes, at page 23, lines 16-20. Thus, the specification teaches that liposomes may be made of neutral lipids, and that the liposome may have a contribution of other lipids. These teachings also support claims to liposomes that "consist essentially of" neutral lipids.

Claims 40, 49 and 53 have been added to describe embodiments of the invention wherein the lipid or the primary phosphatide form a liposome. Support for these claims can be found throughout the specification and claims as filed, with particular written support being found at least at page 23, lines 16-20, and page 4, lines 21-27, which states in relevant part:

> A polynucleotide associated with a lipid may be encapsulated in the aqueous interior of a liposome, interspersed within the lipid bilayer of a liposome, attached to a liposome via a linking molecule that is associated with both the liposome and the polynucleotide...

Additional written support for lipids forming liposomes can also be found at page 5, lines 1-7; page 5, lines 20-21; page 6, lines 7-11; page 6, lines 22-23; page 21, lines 14-20; page 22,

lines 5-16; page 22, line 22 to page 23, line 3; page 23, line 4 to page 25, line 23; page 33, lines 11-20; and originally filed claims 5 and 14.

Claim 52 has been added to describe an embodiment wherein the lipid associated with the BCL-2 oligonucleotide is a "primary phosphatide". Support for claim 52 can be found throughout the specification and claims as filed, at least at page 23, lines 16-20:

> Phospholipids from natural sources, such as egg or soybean phosphatidylcholine, brain phosphatidic acid, brain or plant phosphatidylinositol, heart cardiolipin and plant or bacterial phosphatidylethanolamine are preferably not used as the primary phosphatide, *i.e.*, constituting 50% or more of the total phosphatide composition, because of the instability and leakiness of the resulting liposomes.

It will be understood that no new matter is included within any of the amended or added claims.

C. Rejection of Claims 21-30 Under 35 U.S.C. § 112, Second Paragraph

The Action rejects claims 21-30 as being vague and indefinite for the use of the term "oligonucleotide nucleotide". Applicants respectfully traverse.

Written support for the term "nucleotide" can be specifically found at page 12, lines 3-5 of the specification. This passage describes "nucleotide" as the degree of complementary bases that may be found in a sequence. However, the basis of this rejection has been rendered moot, as claims 21-30 have been revised to even more distinctly claim the invention as an "oligonucleotide".

Applicants respectfully request that this rejection be withdrawn.

D. Rejection of Claims 1-9 and 31-37 Under 35 U.S.C. § 103(a) Over Evan or Reed or Green *et al.* in view of Tari *et al.*

Claims 1-9 and 31-37 are rejected under 35 U.S.C. § 103(a) as allegedly being obvious in light of Evan (WO 93/20200) or Reed (WO 95/08350) or Green *et al.* (U.S. Patent No. 5,583,034) in view of Tari *et al.* (U.S. Patent No. 5,417,978), maintaining the rejection made in the previous Action mailed March 16, 1998. Applicants respectfully traverse.

Tari *et al.* teaches both charged and uncharged lipids, and yet does not teach or suggest a BCL-2 oligonucleotide or a P-ethoxy oligonucleotide. Furthermore, there is no mention of neutral lipids in Evan, Reed or Green *et al.* These references, either alone or in combination, do not teach or suggest that neutral lipids should be employed to produce BCL-2 oligonucleotide/neutral lipid associations. The existence of superior, surprising and unexpected properties of the claimed compositions, namely its specific toxicity to target cells without toxicity to non-target cells relative to charged liposome constructs, argues strongly in favor of a finding of non-obviousness. Moreover, without a teaching or suggestion to associate a BCL-2 oligonucleotide with a neutral lipid to form the BCL-2/neutral lipid constructs of the present invention, there is no motivation to combine these references.

1. Tari *et al.* Does Not Teach or Suggest the Claimed Invention

The Action takes the position that Tari *et al.* teaches that "the liposome is made from a neutral phospholipid selected from a phosphatidylcholine or a phosphatidylserine". This is incorrect. Phosphatidylserine is a negatively charged (*i.e.* anionic) lipid. Although phosphatidyl cholines are neutral and described as being preferred, charged phospholipids, specifically

phosphatidyl serines, are also described as preferred lipids, at column 2, lines 10-13. Tari *et al.* additionally describes liposomes as including other phospholipids, at column 3, lines 41-48:

> "Liposomes" is used in this patent to mean lipid-containing vesicles having a lipid bilayer, as well as other lipid carrier particles which can entrap antisense oligonucleotides. The liposomes can be made of one or more phospholipids, optionally including other materials such as sterols. Suitable phospholipids include phosphatidyl cholines, phosphatidyl serines, and many others that are well known in this field. (Emphasis added.).

This teaching of both charged and uncharged phospholipids as preferred, "and many others that are well known in the art", is not the same as teaching "a neutral lipid" for the construction of liposomes. This reference's teaching of both charged and uncharged lipids and oligonucleotides other than an antisense BCL-2 oligonucleotide does not suggest the claimed combination of neutral lipids with BCL-2 polynucleotides. Applicants submit that, given Tari *et al.*'s teaching of both charged and uncharged lipids, the teaching of countless oligonucleotides without a specific teaching or suggestion of BCL-2 oligonucleotides or P-ethoxy oligonucleotides, and the complete lack of guidance to select a BCL-2 specific polynucleotide, this reference fails to provide the motivation to select the claimed combination of neutral lipid liposomes and a BCL-2 specific polynucleotide.

The Claimed Composition has Surprising, Unexpected, and Superior Properties

Tari *et al.* does not teach or suggest that neutral lipids are selectively less toxic when combined with BCL-2 oligonucleotide, or that charged phospholipids are toxic to cells and thus should be avoided in the case of BCL-2 antisense molecules. Applicants submit that either "superiority in a property" or "the presence of a property not possessed by the prior art" or the "absence of an expected property" is evidence of unobviousness (M.P.E.P. 716.02(a)). This

superiority in the properties of selective toxicity to target cells and non-toxicity to other cell types is demonstrated in the specification at page 7, lines 12-16 and Fig. 1, by the results showing that liposomes comprising neutral lipids and a BCL-2 oligonucleotide are selectively toxic to cells containing a t(14;18) translocation. This property is further demonstrated in the results showing that liposomes comprising neutral lipids with or without a non-BCL-2 oligonucleotide are relatively non-toxic to either target or control cell lines, at page 7, lines 17-22 and in Fig. 2. The evidence of the non-toxicity of liposomes comprising neutral lipids is further described at page 37, lines 3-8:

> Two control oligonucleotides were used to determine the specificity of the inhibition observed. When L-control oligos or empty liposomes were added to Johnson cells, cell growth inhibition was not observed. Jurkat, Raji and Daudi cells were also treated with L-control oligos and empty liposomes. Non-specific toxicity could be observed when greater than 6 μmol/L of L-OS were used, but not with empty liposomes (FIG. 2). (Emphasis added).

The executed declaration provided by Drs. Tari and Lopez-Berestein with the papers submitted June 15, 1998 provides further support of the surprising and unexpected properties of the claimed invention. This declaration demonstrates, in the table at page 3, the selective cytotoxicity of neutral liposomes when combined with a BCL-2 antisense oligonucleotide, the lack of cytotoxicity of neutral (*i.e.* DOPC) liposomes containing a control BCL-2 oligonucleotide, and the non-specific cytotoxicity of liposomes containing 30% negatively charged lipids (DOPC-DMPG) or 30% positively charged lipids (DOPC-DC-CHOL) with either BCL-2 antisense or control oligonucleotides.

These results are also surprising and unexpected in showing that liposomes should be essentially neutrally charged when comprising a BCL-2 antisense oligonucleotide associated with neutral lipids to avoid non-specific toxicity that occurs when charged lipids comprise at

least 30% of the liposome. The teachings of the specification and the evidence provided by this declaration compels a finding of non-obviousness of the claimed invention over this reference which teaches both neutral and negatively charged lipids without any teaching or suggestion of these properties, or a BCL-2 oligonucleotide or P-ethoxy oligonucleotide, or any subsequent guidance in how to create the claimed neutral lipid/BCL-2 oligonucleotide associations.

Further, Applicants submit that the issues here are similar to those found in *In re May*, 574 F.2d 1082, 197 U.S.P.Q. 611 (CCPA 1978). In *In re May*, the court stated:

> "the basis of the prima facie case of obviousness, at least to a major extent, is based on the presumed expectation that compounds which are similar structure will have similar properties. The Wilder II court recognized that a showing of actual difference in properties between the claimed compound and the structurally similar prior art compound over which it was rejected is *not* the only manner of rebutting this presumption...an applicant may rebut the aforementioned presumption by producing sufficient evidence which demonstrates a substantial degree of unpredictability in the pertinent art area." (Citations omitted, emphasis added).

The evidence provided by the declaration that demonstrates a neutral liposome combined with BCL-2 oligonucleotide has selective toxicity to target cells compared to compositions that differ by the presence 30% negatively or positively charged phospholipids, shows the high degree of unpredictability in this field of art. The claimed compositions are not obvious in view of the teachings of Tari *et al.* due to this large degree of unpredictability in the biological properties of BCL-2 oligonucleotide/neutral lipid associations.

The Dependent Claims are Further Distinguished from Tari et al.

Claims 39, 41, 44, 46, 48, 50 and 54 have been added to describe embodiments of the invention wherein the liposome "consists essentially of neutral lipids." This transition language

further clarifies the novel and non-obvious features of the claimed composition by embracing liposomes that are

> the specified material or steps "and those that do not materially affect the basic and novel characteristics(s)" of the claimed invention

MPEP 2111.03, *In re Herz*, 537 F.2d 549, 551-52, 1990 USPQ 461, 463 (CCPA 1976)(emphasis in the original). In this case, the specified material is neutral lipids. The novel and nonobvious features of the claimed invention reside in the surprising, unexpected and superior properties of liposomes made of neutral lipids and BCL-2 antisense oligonucleotides over similar compositions that are non-specifically toxic due to the presence of substantial amounts of charged lipids. Tari *et al.* teaches both charged and uncharged lipids may be used to make liposomes, with no mention of the BCL-2 oligonucleotides or the novel and non-obvious properties of neutral lipid liposome/BCL-2 complexes.

Claims 38, 42, 43, 45, 47, 51 and 55 have been added to describe embodiments of the invention wherein the polynucleotide or oligonucleotide is a P-ethoxy oligonucleotide. These embodiments of the invention are further distinguished from Tari *et al.* by the selection of P-ethoxy oligonucleotides which are "preferred because they are stable in serum and effectively transported into the cellular cytoplasm" (page 25, lines 14-15). Tari *et al.* teaches "methyl phosphonate derivatives of antisense oligonucleotides" at column 3, lines 23-40. Tari *et al.* makes no mention of P-ethoxy oligonucleotides, or BCL-2 oligonucleotides, or BCL-2 oligonucleotides that are P-ethoxy oligonucleotides. It is the instant application, however, that teaches both that BCL-2 oligonucleotides in neutral lipid liposomes, a composition that has surprising and unexpected properties, and P-ethoxy oligonucleotides which possess preferred properties of stability and improved cytoplasmic transport.

2. Evan, Reed and Green *et al.* Do Not Teach or Suggest the Claimed Invention

Evan, Reed and Green *et al.* are purported to teach the use of an antisense oligonucleotide targeted to BCL-2, which is preferably delivered into cells employing liposomes. The present Action admits that none of these references teach a liposome made of neutral lipids, and maintains the rejection for the reasons of record in the previous Action mailed March 16, 1998. The rejection made in the previous Action mailed March 16, 1998, admits at page 11 that "none of these references teach a liposome made of neutral lipids, phosphatidylcholine, phosphatidylglycerol, phophatidylethanolamine or dioleoylphosphatidylcholine."

Evan, Reed or Green *et al.* do not teach or suggest neutral lipids associated with a BCL-2 oligonucleotide. Thus, without this teaching or guidance, these references thus do not make obvious the claimed invention in light of the surprising and unexpected results described above for compositions comprising neutral lipids associated with BCL-2 oligonucleotides.

3. Evan, Reed, Green *et al.* and Tari *et al.* Do Not Teach or Suggest the Claimed Invention, and Are Not Properly Combined

These references, if combined, do not teach or suggest the claimed compositions, as they fail to teach or suggest a BCL-2 oligonucleotide/neutral lipid association. Evan, Reed, Green *et al.* and Tari *et al.* do not teach or suggest the claimed neutral lipid BCL-2 compositions. Moreover, Tari *et al.*, which is relied upon by the Action for a teaching or suggestion of neutral lipids instead describes both neutral and anionic lipids as preferred, and does not teach or suggest that neutral lipids should be combined with BCL-2 oligonucleotides either alone or when combined with the other references.

Applicants further submit that the PTO must show some suggestion in the references themselves or in the knowledge generally available to one of skill in the art for the combining of two or more prior art references. *In re Fine*, 837 F.2d 1071, 1074, 5 U.S.P.Q.2d 1596, 1598-99; Fed. Cir. 1988. The motivation to combine the teachings of Evan, Reed, Green *et al.* with Tari *et al.* described at page 5 of the Action is based on an incorrect premise. The Action is incorrect in its position that Tari *et al.* teaches "the benefits of using liposomes consisting of neutral lipids...(column 2, lines 49-56)" as this reference describes both anionic and neutral lipids, and that these "benefits" are being taught for liposomes constructed from either charged and uncharged lipids. These "benefits" are also not the same as the benefits of BCL-2 oligonucleotide/neutral lipid liposome constructs. The surprising and unexpected specific toxicity to targeted cells that of the claimed invention possess over charged lipid liposome constructs differentiates it from the compositions that these references seem to teach. It is therefore inappropriate to combine these references with Tari *et al.* in an attempt to establish a *prima facie* case of obviousness. No suggestion for doing so in the references themselves or in the generally available knowledge in the art prior to the present invention. *Ex parte Clapp*, 227 U.S.P.Q. 972 (B. P.A.I. 1985).

In light of the foregoing, Applicants respectfully request that this rejection be withdrawn.

E. Rejection of Claims 1-3, 5-8, 10-31 and 33-36 Under 35 U.S.C. § 103(a) over Abubakr *et al.*, Pocock *et al.* and Cotter *et al.* in view of Tari *et al.*

Claims 1-3, 5-8, 10-31 and 33-36 are rejected under 35 U.S.C. § 103(a) over Abubakr *et al.* (*Blood* 82 (10 Suppl. 1) 374a, Abstract #1481), Pocock *et al.* (*Blood* 82 (10 Suppl. 1, 200A, Abstract #784) and Cotter *et al.* (*Oncogene*, 9:3049-3055, 1994) as allegedly being obvious in view of Tari *et al.* (U.S. Patent No. 5,417,978). Applicants respectfully traverse.

Tari *et al.* does not teach or suggest the claimed invention, as described in section D.

The Action admits that neither Abubakr *et al.*, Pocock *et al.* or Cotter *et al.* teach the administration of an antisense oligonucleotide as a composition comprising neutral lipids. Applicants have not found in either Abubakr *et al.*, Pocock *et al.* or Cotter *et al.* any mention or teaching of any lipid, let alone a neutral lipid, for combining with an antisense BCL-2 oligonucleotide. These references are therefore irrelevant to the claimed invention, either when considered alone or combined with each other, or with Tari *et al.*, to establish a *prima facia* case of obviousness.

The Action argues that one of skill in the art would be "motivated by the teaching of Tari *et al.* that the neutral lipid compositions imparts several benefits on the administration of an antisense oligonucleotide." As described in section D of this paper, Tari *et al.* teaches the benefits of both anionic and neutral lipids. The premise for the motivation to combine is incorrect, and Applicants submit it is inappropriate to combine these references without some suggestion in the references themselves or in the knowledge generally available to one of skill in the art for the combining of two or more prior art references. *In re Fine*, 837 F.2d 1071, 1074, 5 USPQ2d 1596, 1598-99; Fed. Cir. 1988. No suggestion for doing so in the references themselves or in the generally available knowledge in the art prior to the present invention. Ex parte Clapp, 227 USPQ 972 (B. P.A.I. 1985).

In light of the foregoing, Applicants respectfully request that this rejection be withdrawn.

F. Rejection of Claims 4 and 32 Under 35 U.S.C. § 112(e) or § 103(a) over Abubakr *et al.*, Pocock *et al.* and Cotter *et al.* in view of Tari *et al.* and further in view of Evan

Claims 4 and 32 are rejected under 35 U.S.C. § 103(a) over Abubakr *et al.* (*Blood* 82 (10 Suppl. 1) 374a, Abstract #1481), Pocock *et al.* (*Blood* 82 (10 Suppl. 1, 200A, Abstract #784) and Cotter *et al.* (*Oncogene*, 9:3049-3055, 1994) as allegedly being obvious in view of Tari *et al.* (U.S. Patent No. 5,417,978) and in further view of Evan (WO 93/20200). Applicants respectfully traverse.

Tari *et al.* does not teach or suggest the claimed invention, as described in section D. Abubakr *et al.*, Pocock *et al.* or Cotter *et al.* do not teach or suggest the claimed invention, as described in section E. The inclusion of Evan does not make the invention of claims 4 and 32 obvious, as the primary references do not teach or suggest the claimed invention of the independent claims from which these claims depend.

As described in section E above, it is inappropriate to combine the primary references. Applicants find no teaching or suggestion in Evan that the primary references should be combined, or a teaching or suggestion in Evan to combine the teachings of this reference with the primary references.

In light of the foregoing, Applicants respectfully request that this rejection also be withdrawn.

G. Conclusion

This is a complete response to the referenced Official Action. In conclusion, Applicants submit that, in light of the foregoing remarks, the present case is in condition for allowance and such favorable action is respectfully requested. Should Examiner Schwartzman have any

questions or comments, or believe that certain actions might more readily progress this case towards allowance, a telephone call to the undersigned Applicants' representative is earnestly solicited.

Respectfully submitted,

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